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Evaluation of the enantiomers of 1-octen-3-ol and 1-octyn-3-ol as attractants for mosquitoes associated with a freshwater swamp in Florida, U.S.A.

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Abstract. Field studies were conducted at wooded wetlands in Gainesville, FL, U.S.A., to assess responses of natural populations of adult mosquitoes (Diptera: Culicidae) to American Biophysics MM-X™ and Coleman MD-2500™ traps baited with enantiomers of 1-octen-3-ol, a naturally occurring compound, and 1-octyn-3-ol, a closely related synthetic compound. Overall, the same species of mosquitoes were attracted by all enantiomers, although the (R)-(+) isomer of octenol generally attracted more species, and it is the isomer produced in greatest proportion in nature. Traps baited with the R-enantiomer caught greater numbers of mosquitoes than those baited with the S-enantiomer of each compound, whereas traps baited with S-enantiomers were equally or slightly less attractive than those baited with carbon dioxide only.

Key words. 1-octen-3-ol, 1-octyn-3-ol, baited traps, mosquito attractants.

Introduction

The compound 1-octen-3-ol (hereafter, octenol) is an unsaturated, 8-carbon alcohol ($CH_3[CH_2]_4CHOH.CH_2 = CH_2$), volatile compound that has been isolated from many natural sources, mainly plants and fungi (Dijkstra & Wiken, 1976). It has been identified in volatiles from clover (Honkanen & Moisio, 1963) and alfalfa (Buttery & Kamm, 1980). It is produced by numerous species of moulds (Kaminski *et al*., 1974) and mushrooms (Wurzenberger & Grosch, 1982), and is produced when grain is contaminated by mould (Kaminski *et al*., 1973). Octenol has also been isolated from both invertebrate and vertebrate animals. Pierce *et al.* (1989) reported that two species of grain beetles, *Oryzaephilus surinamensis* (Linn) and *Oryzaephilus mercator* (Fauvel) (Coleoptera: Silvanidae), produce octenol that serves as an aggregation pheromone. Octenol has been reported by several investigators as a mammalian emanation (Hall *et al.*, 1984; Raymer *et al.*, 1985), but it has not been reported in ornithophilic emanations. Hall *et al.* (1984) found octenol to be present in the breath of oxen. Raymer *et al.* (1985) reported that octenol is apparently produced by micro-organisms in the anal sac of wolves. Octenol is also present in low concentrations in human sweat and it activates mosquito antennal receptors (Cork & Park, 1996).

Octenol occurs naturally as two optically active enantiomers: (S)-(+) and R-(−), with carbon-3 as the chiral centre. The ratio of the enantiomers varies according to the source, but R-octenol seems to be the predominant enantiomer (Dijkstra & Wiken, 1976; Hall *et al.*, 1984; Pierce *et al.*, 1989). Both isomers have been shown to be attractive to grain beetles (Pierce *et al.*, 1989) and tsetse (Hall *et al.*, 1984). In different samples of ox odour the enantiomeric R : S composition varied from 80 : 20 to 92 : 8 (Hall *et al.*, 1984). When presented singly in odour-baited traps, the two enantiomers were equally effective in capturing tsetse. In most experimental studies with octenol, investigators have used the racemic mixture.

Octenol is a proven kairomone used by a variety of haematophagous insects to locate their vertebrate hosts; first demonstrated for tsetse, *Glossina* (*Glossina*) *morsitans* (Westwood) (Diptera: Glossinidae) by Hall *et al.*, 1984. Takken & Kline (1989) were first to report that octenol also serves as a kairomone for several mosquito species. Since then, numerous studies have confirmed that octenol is an attractant for many, but not all, mosquito genera and species (Kline *et al.*, 1990a, b, 1991a, b; Kemme *et al.*, 1993; Becker *et al.*, 1995). Several studies demonstrated that octenol, as a supplement to carbon dioxide (CO_2), significantly increased collections of *Aedes* but not *Culex* mosquitoes. Octenol has also shown evidence of attraction in

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traps for zoophilic species of Muscidae (Cork, 1994), Tabanidae (French & Kline, 1989; Hayes *et al*., 1993) and Ceratopogonidae (Kline *et al.*, 1994), as well as some species of Simuliidae (Cheke & Garms, 1987; Atwood & Meisch, 1993). Gibson & Torr (1999) pointed out that there was no evidence of a response in phlebotomine sandflies; however, electrophysiological studies by Sant'Ana *et al.* (2002) demonstrated that *Lutzomyia longipalpis* (Lutz & Neiva) antennae possess chemoreceptors for this compound.

We were unable to find any published comparisons of the roles of the two enantiomers of octenol in relation to the host-seeking behaviour of mosquitoes, nor any reported evaluation of 1-octyn-3-ol (hereafter, octynol), a synthetic octenol analogue, as a mosquito attractant. Octynol also occurs as two active enantiomers: (S)-(−) and R-(+) with carbon-3 again being the chiral centre. Therefore, the primary objectives of the experiments reported in this paper were to determine the relative efficacy of octynol compared with octenol, the enantiomers of each compound, and various blends of the enantiomers of both compounds for trapping mosquitoes from natural populations in Florida.

As most previous studies with octenol as a mosquito kairomone demonstrated only a slightly increased response to octenol alone, but a synergistic increase to the combination of octenol and CO_2 (Takken & Kline, 1989; Kline *et al.*, 1990a, 1991a, b; Kemme *et al.*, 1993; Van Essen *et al.*, 1994), all our experiments were conducted with traps supplemented with CO_2 from a compressed gas tank or with traps that produced an equivalent amount of CO_2 through the combustion of propane.

Materials and methods

Six field trapping experiments were conducted against natural populations of mosquito species associated with wooded freshwater wetlands located near a water management area in Gainesville, FL. In each experiment a 4 × 4 Latin square experimental design was used. Mosquito lures were either the commercially available standard lure produced by BioSensory, Inc., (Putnam, CT, U.S.A.; www.biosensory.com) or special lures provided by Bedoukian Research Inc. (Danbury, CT, U.S.A.; www.bedoukian.com). The BioSensory lure consisted of 3.75 g of a 50 : 50 R : S racemic blend of octenol formulated in 12 g of a patented blend of waxes, which slowly released the octenol from a patented plastic dispenser. All other octenol and octynol lures were provided by the Bedoukian Research Inc., which utilized the 'BioSensory-type' wax mix and dispensers. Enantiomeric purity was: R-octenol, 99%; S-octenol, 95%; R-octynol, 97.5%, and S-octynol, 96% (R. H. Bedoukian, personal communication). Each lure contained either 2.5 g of the octenol or octynol enantiomers for the single compound lures, or 1.25 g of the desired octenol enantiomers and/or 1.25 g of the desired octynol enantiomers for the mixed lures, in 12 g of wax. Based on research conducted by Kline *et al.* (1991b), all lures were designed to release 50 mg of the attractant per day.

The first two experiments were conducted within a single wooded area (50 × 100 m) located in a suburban residential area. Four trap stations were located at the four corners, so that the minimum distance between stations was 50 m. In these two experiments an MM-X™ trap (American Biophysics Corp. [ABC], North Kingstown, RI, U.S.A.) was used at each trap station. This trap design utilizes a relatively new mosquito-capturing principle known as CounterFlow™ (patented by ABC), first described in detail by Kline (1999). In operation the trap utilizes two fans energized by a 12-V DC battery to provide CounterFlow™ at the trap entrance. A 40-mm fan (Delta model DFB0412M, Delta Electronics Inc., Alexandria, VA) creates a CO_2-enriched airflow plume from CO_2 supplied from a compressed gas cylinder, which exits vertically down a centre pipe. An upflow is created by an 80-mm fan (Delta model DFB0812H, Delta Electronics) that causes any mosquito with a flight speed of less than ~ 3.5 m/sec, when in the vicinity of the trap entrance, to be entrained with the upflow and forced into the trap interior. The MM-X trap was hung from a pole so that the bottom of the attractant plume was ~ 50 cm above the ground. Based on previous field studies with a variety of mosquito species (Kline *et al.* 1990a, b, 1991a, b), CO_2 was supplied to each MM-X trap at 500 mL/min from 9-kg compressed gas cylinders through polyethylene tubing. Control of the CO_2 flow rate was achieved with FLOWSET1 (ABC), which consists of a pressure regulator with output fixed at 1.06 kg/cm^2, a 10-μm line filter, a 500-mL/min flow control orifice (0.007), and quick connect lure fittings.

The treatment locations were randomly chosen for the first night. On each subsequent night the treatments were rotated counter-clockwise to the next location, so that each treatment occupied each position only once during the four-night experiment.

The first experiment was conducted during 13–17 July 2002 (4 nights, no replicates). Traps were baited with one of four treatments: 500 mL/min CO_2 only; 500 mL/min CO_2 + a Bedoukian-produced (BioSensory-type) racemic octenol lure (50 : 50, R : S octenol enantiomers); 500 mL/min CO_2 + R-octenol lure, and 500 mL/min CO_2 + S-octenol lure.

In the second experiment, conducted during 22–26 August 2002 (4 nights, no replicates), the treatments were: 500 mL/min CO_2 only; 500 mL/min CO_2 + a Bedoukian R : S racemic octynol lure; 500 mL/min CO_2 + R-octynol lure, and 500 mL/min CO_2 + S-octynol lure. Lures were suspended from the bottom of the MM-X traps with twist ties. Traps were set each day between 08.00 and 09.00 hours and operated for ~ 24 h. Traps were replaced each day with new traps. Traps removed from the field were placed in a walk-in cold room to immobilize the mosquitoes, which were then transferred into containers and stored in a freezer until they were processed for counting and identification of species.

Four more experiments, conducted in the same suburban residential area, employed MD-2500™ traps (Coleman Co., Wichita, KS, U.S.A.), which utilize combustion of propane from standard 9-kg cylinders to produce warm, moist CO_2 at the rate of ~ 500 mL/min. A single trap was placed in each of four different fields, which were not adjacent to each other. Traps were > 100 m apart and operated continuously. Treatments were changed daily between 08.00 and 09.00 hours. Mosquitoes were collected in the trap net located in the trap's collection box. The entire collection box from each trap was returned to the laboratory each day and placed in a freezer. The contents of each net were counted and identified to species. Treatments consisted either of the commercially available BioSensory standard R : S racemic octenol lure or the specially formulated lures provided by Bedoukian Research Inc.

The third experiment, conducted during 9–27 March 2005 (16 nights, four replicates), compared four treatments: CO_2 only (releasing CO_2, heat and water vapour produced by the combustion of propane, but no additional lures); CO_2 + R-octenol; CO_2 + S-octenol, and CO_2 + R-octynol. The fourth experiment, conducted during 9–13 June 2005 (4 nights, no replicates), compared: CO_2 only; CO_2 + R-octenol; CO_2 + R-octynol, and CO_2 + 50 : 50 mix of R-octenol : R-octynol. The fifth experiment was conducted during 15 July–16 August 2005 (four replicates) and compared: CO_2 only; CO_2 + R-octynol; CO_2 + S-octynol, and CO_2 + a commercially available standard R : S racemic octenol lure (BioSensory, Inc.). The sixth experiment, conducted during 28 April–18 May 2005 (four replicates), compared: CO_2 + standard R : S racemic octenol BioSensory lure; CO_2 + R-octynol; CO_2 + R-octenol, and CO_2 + 50 : 50 mix of R-octenol : R-octynol.

Species identification employed the keys of Darsie & Ward (2005). Trap counts of each species were transformed to log $(n + 1)$ and the data analysed with the Statistical Analysis System (SAS) PROC ANOVA (SAS Institute, 2001) to determine the effect of day, location and treatment on the number of mosquitoes of each species trapped. Comparisons of means were performed on transformed data with SAS PROC MEANS/Tukey (SAS Institute, 2001) but detransformed for the tables in the Results section.

Results

The first experiment with MM-X traps (Table 1) yielded a total of 3566 mosquitoes (all females) of 17 species from four traps operated for four nights. The largest numbers of mosquitoes were collected by R : S racemic octenol + CO_2 traps (1310), followed by R-octenol + CO_2 traps (1250), S-octenol + CO_2 traps (652) and CO_2-only traps (354). The following species were collected (in descending order of abundance): 3103 *Ochlerotatus infirmatus* Dyar & Knab (also known as *Aedes infirmatus*); 165 *Psorophora ferox* (Von Humboldt); 93 *Anopheles crucians* Wiedemann; 39 *Ochlerotatus triseriatus* (Say) (also known as *Aedes triseriatus*); 32 *Culex erraticus* (Dyar & Knab); 31 *Coquillettidia perturbans* (Walker); 20 *Culex nigripalpus* Theobald; 18 *Aedes vexans* (Meigen); 18 *Ochlerotatus fulvus-pallens* Ross (also known as *Aedes fulvus-pallens*); 16 *Aedes albopictus* (Skuse); 11 *Culex salinarius* Coquillett; 11 *Anopheles quadrimaculatus* Say; three *Ochlerotatus taeniorhynchus* Wiedemann (also known as *Aedes taeniorhynchus*); two *Psorophora ciliata* (Fabricius); two *Psorophora howardii* Coquillett; one *Culex quinquefasciatus* Say, and one *Ochlerotatus atlanticus* (Dyar & Knab) (also known as *Aedes atlanticus*). Among these, 14 species were collected by R-octenol, 14 by S-octenol, 13 by R : S racemic octenol and 13 by CO_2 only. Species not collected with R-octenol were *Cx. quinquefasciatus*, *Oc. atlanticus* and *Ps. howardii*; those not collected with S-octenol were *Cx. quinquefasciatus*, *Oc. atlanticus* and *Ps. ciliata*; those not collected with R : S racemic octenol were *Oc. atlanticus*, *Oc. taeniorhynchus*, *Ps. ciliata* and *Ps. howardii*, and those not collected by CO_2 only were *Cx. quinquefasciatus*, *Ps. ciliata*, *An. quadrimaculatus* and *Ps. howardii*.

As shown in Table 1, the mean total mosquito collections in traps baited with R-octenol + CO_2 and R : S racemic octenol + CO_2 were > three-fold greater than and significantly different ($P < 0.05$) to collections from traps baited with CO_2 only, but not to those of traps baited with S-octenol + CO_2. The mean catch in traps baited with S-octenol + CO_2 was not significantly greater (although 1.84-fold) than that of traps baited with CO_2

Table 1. Mean catch (± standard error) per trap-day* of most frequently caught species for different treatments of odour-baited MM-X traps (all traps baited with 500 mL/min CO_2); Gainesville, FL; 13–17 July 2002.

Species	Attractant			
	CO_2 only	S-octenol	R-octenol	R : S octenol
Anopheles crucians	0.7 (0.25)	6.7 (2.36)	9.5 (1.94)	6.2 (2.84)
Anopheles quadrimaculatus	0.0 (0.0)	0.5 (0.50)	1.2 (0.48)	1.0 (0.58)
Aedes albopictus	0.7 (0.48)	1.0 (0.71)	1.0 (0.00)	1.2 (0.25)
Aedes vexans	1.2 (0.63)	0.5 (0.29)	1.5 (0.87)	1.2 (0.48)
Culex erraticus	0.7 (0.48)	3.5 (2.84)	2.2 (1.11)	1.5 (0.50)
Culex salinarius	0.2 (0.25)	0.5 (0.29)	0.2 (0.25)	1.7 (0.85)
Culex nigripalpus	2.0 (0.71)	0.7 (0.25)	0.7 (0.75)	1.5 (0.87)
Coquillettidia perturbans	0.5 (0.50)	1.0 (0.41)	2.2 (0.75)	4.0 (2.71)
Ochlerotatus infirmatus	74.5 (22.53)B	136.7 (14.05)AB	271.7 (71.49)A	292.7 (74.12)A
Ochlerotatus triseriatus	1.5 (0.65)	1.5 (0.96)	3.0 (0.71)	3.7 (2.78)
Ochlerotatus fulvus-pallens	0.5 (0.5)	0.5 (0.50)	2.2 (1.11)	1.2 (0.75)
Psorophora ferox	5.2 (1.89)	9.0 (2.74)	16.0 (6.56)	11.0 (1.08)
Total	88.5 (23.24)B	163.0 (13.25)AB	312.5 (72.89)A	327.5 (80.29)A

*$n = 4$ trap-days per treatment; means in the same row followed by different letters are significantly different ($P < 0.05$); Tukey's means separation applied to log $(n + 1)$ transformed data.

only, and not significantly less (~ two-fold) than collections with the other enantiomers. These contrasts between trap treatments were significant for *Oc. infirmatus*, the most abundant species, but *Cx. nigripalpus* was not affected by any of the octenol baits. Overall, Experiment 1 shows that R-octenol and R : S racemic octenol are attractants for total mosquitoes caught and for the most abundant species (*Oc. infirmatus*), and S-octenol is probably an attractant, but only mildly so.

In the second experiment with MM-X traps (Table 2), two male as well as 5985 female mosquitoes of 24 species were collected from the four traps operated for four nights. Traps baited with R : S racemic octynol + CO_2 caught the largest numbers (1724 ♀, 1 ♂), followed by the R-octynol + CO_2 treatment (1678 ♀), CO_2 only (1430 ♀) and S-octynol + CO_2 (1155 ♀, 1 ♂). In descending order of abundance, species collected were: 2903 ♀ *Oc. infirmatus*; 2242 ♀, 1 ♂ *Cx. nigripalpus*; 200 ♀ *Oc. fulvus-pallens*; 181 ♀ *An. crucians*; 118 ♀ *Ps. ferox*; 111 ♀ *Cq. perturbans*; 64 ♀ *Oc. atlanticus*; 59 ♀ *Cx. erraticus*; 33 ♀ *An. quadrimaculatus*; 21 ♀ *Ae. albopictus*; 18 ♀ *Psorophora columbiae*; 11 ♀ *Oc. triseriatus*; 5 ♀ *Ochlerotatus dupreii* (Coquillett); 4 ♀ *Cx. salinarius*; 1 ♂, 3 ♀ *Ps. howardii*; 3 ♀ *Ae. vexans*; 3 ♀ *Psorophora ciliata*, 2 ♀ *Ochlerotatus canadensis* (Theobald), and single females of *Anopheles punctipennis* (Say), *Cx. quinquefasciatus*, *Mansonia titillans* (Walker), *Uranotaenia lowi* Theobald, *Uranotaenia sapphirina* (Osten Sacken) and *Ochlerotatus mitchellae* (Dyar). Species not collected with R-octynol were *Ae. vexans*, *Cx. quinquefasciatus*, *Ma. titillans*, *Oc. canadensis*, *Oc. mitchellae*, *Ur. lowi* and *Ps. howardii*; those not collected with S-octynol were *Ae. vexans*, *An. punctipennis*, *Cx. salinarius*, *Oc. triseriatus*, *Cx. quinquefasciatus*, *Ma. titillans*, *Ur. lowi*, *Ur. sapphirina* and *Ps. ciliata*; those not collected with R : S racemic octenol were *An. punctipennis*, *Cx. salinarius*, *Ma. titillans*, *Ur. lowi*, *Ur. sapphirina*, *Oc. dupreii*, *Oc. canadensis*, *Ps. ciliata* and *Oc. mitchellae*, and those not collected with CO_2 only were *An. punctipennis*, *Cx. quinquefasciatus*, *Ur. sapphirina*, *Ps. ciliata*, *An. quadrimaculatus*, *Oc. mitchellae* and *Ps. howardii*.

As shown in Table 2, there were no significant differences between the catches in any of the octynol-baited traps and the CO_2-only trap, for the mean total catches or for individual species, although there were consistent trends for the most abundant species; R : S racemic octynol + CO_2 or R-octynol + CO_2 caught slightly more (1.2-fold), whereas S-octynol + CO_2 caught slightly less (0.8-fold) mosquitoes than traps baited with CO_2 only. No such pattern was evident for the most abundant *Culex* species.

The third experiment, this time employing Coleman traps, yielded a total of 4698 mosquitoes of 15 species over 16 nights (Table 3). Traps baited with R-octenol + CO_2 collected the most (2222 ♀, 39 ♂), followed by traps baited with R-octynol + CO_2 (1484 ♀, 10 ♂). Traps baited with S-octenol + CO_2 (495 ♀, 6 ♂) and CO_2 only (494 ♀, 8 ♂) collected nearly the same number of mosquitoes. In descending order of abundance, species collected were: 2789 ♀ *An.* crucians; 1242 ♀, 55 ♂ *Cx. salinarius*; 358 ♀ *Cx. erraticus*; 126 ♀ *Oc. canadensis*; 62 ♀ *Cx. nigripalpus*; 56 ♀, 1 ♂ *Culiseta melanura* (Coquillett); 22 ♀ *Oc. infirmatus*; 11 ♀ *Oc. atlanticus*; 10 ♀ *An. quadrimaculatus*; 6 ♀, 7 ♂ *Culex territans* Walker; 6 ♀ *Cq. perturbans*; 3 ♀ *Ps. ferox*; 2 ♀ *An. punctipennis*; 1 ♀ *Oc. taeniorhynchus* and 1 ♀ *Oc. triseriatus*. Species not collected with R-octenol were *Oc. taeniorhynchus* and *Oc. triseriatus.* Those not collected with S-octenol were *Oc. atlanticus*, *Oc. taeniorhynchus* and *Oc. triseriatus.* All but *Oc. triseriatus* were collected with R-octynol. Those not collected in the CO_2-only trap were *An. punctipennis*, *Oc. atlanticus*, *Oc. infirmatus*, *Oc. taeniorhynchus* and *Ps. ferox*.

As shown in Table 3, the mean total mosquito collection of traps baited with R-octenol + CO_2 was significantly greater (nearly five-fold) than that of CO_2-only traps and of traps baited with S-octenol + CO_2, but not significantly greater (1.5-fold) than that of R-octynol + CO_2-baited traps, which yielded a collection intermediate in size to those of the other treatments.

Table 2. Mean catch (± standard error) per trap-day* for most frequently caught species for different treatments of odour-baited MM-X traps (all traps baited with 500 mL/min CO_2); Gainesville, FL; 22–26 August 2002.

	Attractant			
Species	CO_2 only	R : S octynol	R-octynol	S-octynol
Anopheles crucians	10.0 (4.02)	15.0 (3.34)	12.0 (1.73)	8.2 (2.72)
Anopheles quadrimaculatus	2.0 (0.58)	2.7 (1.03)	1.0 (0.41)	2.5 (1.04)
Aedes albopictus	1.7 (1.03)	0.5 (0.29)	0.7 (0.25)	2.2 (1.60)
Culex nigripalpus	142.5(45.93)	131.5 (41.32)	149.7 (66.13)	136.7 (29.77)
Culex erraticus	3.2 (1.65)	4.7 (1.80)	2.7 (1.44)	4.0 (1.47)
Coquillettidia perturbans	4.7 (1.93)	10.5 (3.71)	8.2 (2.98)	4.2 (1.80)
Ochlerotatus atlanticus	4.2 (2.29)	5.7 (2.21)	4.7 (1.54)	1.2 (0.63)
Ochlerotatus infirmatus	166.2 (57.67)	230.5 (66.18)	213.0 (59.39)	116.0 (27.74)
Ochlerotatus triseriatus	0.5 (0.50)AB	0.5 (0.5)AB	1.7 (0.25)A	0.0 (0.00)B
Ochlerotatus fulvus-pallens	12.0 (5.58)	18.0 (8.13)	14.2 (6.42)	5.7 (1.80)
Psorophora columbiae	0.5 (0.29)	1.5 (1.50)	1.0 (0.58)	1.5(0.87)
Psorophora ferox	6.7 (2.56)	9.0 (3.19)	8.7 (2.81)	5.0 (1.69)
Total	357.5 (24.97)	431.0 (77.0)	419.5 (96.36)	288.7 (18.47)

*$n = 4$ trap-days per treatment; means in the same row followed by different letters are significantly different ($P < 0.05$); Tukey's means separation applied to log ($n + 1$) transformed data.

Table 3. Mean catch (± standard error) per trap-day* of most frequently caught species for different treatments of odour-baited Coleman MD-2500 propane-powered traps (traps produced ~ 500 mL/min CO_2, heat and water vapour), Gainesville, FL; four replicates of a 4 × 4 Latin square experiment; 9–27 March 2005.

	Attractant			
Species	R-octenol	S-octenol	R-octynol	CO_2 only
Anopheles crucians	93.4 (30.06)A	14.6 (4.51)BC	53.6 (18.33)AB	12.5 (4.36)C
Culex salinarius	31.0 (6.82)A	10.5 (2.36)B	24.2 (5.40)AB	11.8 (2.83)AB
Culex nigripalpus	1.5 (0.80)	0.5 (0.22)	1.2 (0.54)	0.5 (0.24)
Culex erraticus	7.2 (2.22)	2.7 (0.94)	8.4 (3.38)	4.0 (1.22)
Culiseta melanura	1.0 (0.47)	1.2 (0.62)	0.4 (0.27)	0.8 (0.32)
Ochlerotatus canadensis	3.3 (1.93)	0.8 (0.36)	2.9 (1.17)	0.8 (0.31)
Total	138.8 (37.28)A	30.9 (7.39)B	92.7 (25.57)AB	30.8 (7.27)B

*$n = 16$ trap-days per treatment; means in the same row followed by the same letter are not significantly different ($P > 0.05$); Tukey's means separation applied to log $(n + 1)$ transformed data.

With regard to *An. crucians*, traps baited with R-octenol + CO_2 caught significantly more than those baited with S-octenol + CO_2 or CO_2 only, whereas the R-octynol + CO_2-baited traps yielded intermediate numbers; R-octynol + CO_2-baited traps caught significantly more (4.3-fold) than the CO_2-only traps ($P < 0.05$). Traps baited with R-octenol + CO_2 caught significantly more *Cx. salinarius* than those baited with S-octenol + CO_2, whereas traps baited with the other two treatments (R-octynol + CO_2 or CO_2 only) yielded intermediate numbers. The catch of *Cx. salinarius* with CO_2 only did not significantly differ from that of any of the other treatments. Only *An. crucians* and *Cs. melanura* were more abundant in S-octenol + CO_2-baited traps than in CO_2-only traps.

Overall, Experiment 3 shows that R-octenol is more attractive than R-octynol for total mosquito numbers and for the most abundant species (*An. crucians*). The results support the conclusion that S-octenol is only mildly, if at all, an attractant and these odours are not attractants for the most abundant *Culex* species.

The fourth experiment, also using Coleman traps, yielded the fewest mosquitoes of all, with totals of 1186 ♀ and 4 ♂ mosquitoes, comprising 16 species over 4 nights (Table 4). Traps baited with equal parts of R-octenol + CO_2 and R-octynol + CO_2 yielded the greatest total number (388 ♀), followed by traps baited with R-octenol + CO_2 (351 ♀, 1 ♂), traps baited with R-octynol + CO_2 (283 ♀, 2 ♂) and CO_2-only traps (164 ♀, 1 ♂). In descending order of abundance, species collected were: 404 ♀ *An. crucians*; 180 ♀ *Cx. erraticus*; 120 ♀ *Cx. nigripalpus*; 112 ♀ *Oc. atlanticus*; 110 ♀ *Cq. perturbans*; 110 ♀ *Oc. infirmatus*; 43 ♀, 1 ♂ *Cx. salinarius*; 43 ♀ *Oc. canadensis*; 21 ♀ *Cx. quinquefasciatus*; 18 ♀ *Ps. ferox*; 15 ♀ *Cs. melanura*; 4 ♀ *Oc. triseriatus*; 1 ♀, 3 ♂ *Ps. howardii*; 2 ♀ *An. quadrimaculatus*; 2 ♀ *Ae. vexans*, and 1 ♀ *Orthropodomyia signifera* (Coquillett). All but three of these species (*An. quadrimaculatus*, *Ae. vexans* and *Ps. howardii*) occurred in the CO_2-only traps. Species not collected with the R-octenol : R-octynol mix were *An. quadrimaculatus*, *Ae. vexans* and *Oc. triseriatus*; those not collected with R-octenol were *Ps. howardii* and *Or. signifera*, and those not collected with R-octynol were *An. quadrimaculatus*, *Ae. vexans* and *Or. signifera*.

The fifth experiment yielded a total of 9259 ♀ and 14 ♂ mosquitoes comprising 20 species over 16 nights (Table 5). Traps baited with the standard R : S octenol lure + CO_2 caught the greatest number (3677 ♀, 3 ♂), followed by traps baited with R-octynol + CO_2 (3385 ♀, 1 ♂), CO_2-only traps (1196 ♀,

Table 4. Mean catch (± standard error) per trap-day* of most frequently caught species for different treatments of odour-baited Coleman MD-2500 propane combustion traps (all traps produce ~ 500 mL/min CO_2, heat and water vapour); Gainesville, FL; 9–13 June 2005.

	Attractant			
Species	CO_2 only	R-octenol	R-octynol	50 : 50 mix
Anopheles crucians	5.0 (2.86)	37.7 (17.36)	25.0 (8.39)	33.2 (17.73)
Culex quinquefasciatus	1.2 (0.63)	0.5 (0.29)	2.0 (1.22)	1.5 (0.29)
Culex salinarius	1.0 (0.58)	3.0 (1.47)	4.2 (0.85)	2.5 (1.19)
Culex nigripalpus	10.2 (7.63)	5.2 (1.11)	8.2 (2.21)	6.2 (2.87)
Culex erraticus	10.2 (3.90)	12.2 (7.47)	12.2 (3.33)	10.2 (2.09)
Culiseta melanura	2.0 (0.71)	1.0 (1.0)	0.2 (.25)	0.5 (0.29)
Coquillettidia perturbans	5.5 (0.96)	9.2 (4.44)	5.5 (2.87)	7.2 (2.69)
Ochlerotatus atlanticus	1.5 (0.87)	5.2 (1.93)	4.2 (1.11)	17.0 (15.02)
Ochlerotatus infirmatus	3.2 (0.75)	8.5 (5.2)	4.5 (1.32)	11.2 (5.29)
Total	41.0 (7.08)	87.7 (35.26)	70.7 (8.38)	97.0 (45.14)

*$n = 4$ trap-days per treatment; means in the same row not significantly different ($P > 0.05$); Tukey's means separation applied to log $(n + 1)$ transformed data.

Table 5. Mean catch (± standard error) per trap-day* of most frequently caught species for different treatments of odour-baited Coleman MD-2500 propane-powered traps (traps produced ~ 500 mL/min CO_2, heat and water vapour), Gainesville, FL; four replicates of a 4 × 4 Latin square experiment; 15 July–16 August 2005.

	Attractant			
Species	R-octynol	S-octynol	R : S octenol	CO_2 only
Anopheles crucians	26.3 (12.41)A	3.7 (1.26)B	26.8 (8.54)A	3.3 (1.09)B
Culex quinquefasciatus	0.2 (0.14)	1.0 (0.51)	0.3 (0.15)	0.7 (.39)
Culex nigripalpus	18.3 (4.26)	14.6 (3.49)	18.1 (4.26)	14.8 (3.15)
Culex erraticus	12.1 (1.95)	7.7 (1.61)	17.8 (4.33)	7.0 (1.87)
Culiseta melanura	0.5 (0.20)	1.4 (0.56)	0.7 (0.29)	1.5 (0.61)
Coquillettidia perturbans	45.9 (8.49)A	17.1 (3.31)B	60.3 (8.56)A	21.5 (3.96)B
Ochlerotatus atlanticus	84.1 (18.54)A	12.3 (2.74)B	85.8 (20.52)A	19.4 (5.59)B
Ochlerotatus infirmatus	18.1 (4.82)A	2.8 (0.7)B	13.4 (3.35)A	3.6 (2.06)B
Ochlerotatus triseriatus	1.0 (0.38)	0.2 (0.11)	0.6 (0.29)	0.2 (0.14)
Mansonia titillans	0.1 (0.09)AB	0.1 (0.09)AB	0.6 (0.29)A	0.0 (0.0)B
Psorophora columbiae	0.5 (0.2)AB	0.1 (0.06)B	1.81 (0.7)A	0.1 (0.14)B
Psorophora ferox	2.1 (0.43)A	0.6 (0.15)B	1.38 (0.41)AB	1.5 (0.53)AB
Total	211.4 (39.15)A	62.6 (10.38)B	229.8 (41.18)A	74.63 (15.99)B

*$n = 16$ trap-days per treatment; means in the same row followed by different letters are significantly different ($P < 0.05$); Tukey's means separation applied to log ($n + 1$) transformed data.

1 ♂) and traps baited with S-octynol + CO_2 (1003 ♀, 3 ♂). In descending order of abundance, species collected were: 3227 ♀ *Oc. atlanticus*; 2317 ♀, 2 ♂ *Cq. perturbans*; 1056 ♀, 1 ♂ *Cx. nigripalpus*; 964 ♀ *An. crucians*; 714 ♀ *Cx. erraticus*; 615 ♀ *Oc.* infirmatus; 92 ♀, 8 ♂ *Ps. ferox*; 67 ♀, 1 ♂ *Cs. melanura*; 42 ♀ *Ps. columbiae*; 38 ♀ *Cx. quinquefasciatus*; 35 ♀ *Oc. triseriatus*; 30 ♀ *Oc. dupreii*; 19 ♀ *Cx. salinarius*; 14 ♀ *Ma. titillans*; 9 ♀ *An. quadrimaculatus*; 8 ♀ *Ae. albopictus*; 5 ♀, 2 ♂ *Cx. territans*; 4 ♀ *Oc. canadensis*; 2 ♀ *Ps. howardii*, and 1 ♀ *Ae. vexans*. Species not collected with the standard R : S octenol lure were *Ae. vexans*, *Cx. territans*, *Oc. taeniorhynchus*, *Ps. howardii* and *Ur. lowi*; those not collected with R-octynol were *Oc. taeniorhynchus* and *Ur. lowi*; those not collected with S-octynol were *Ae. vexans*, *Cx. territans*, *Oc. canadensis*, *Oc. taeniorhynchus* and *Ps. howardii*, and those not collected with CO_2 only were *Ae. vexans*, *Cx. territans*, *Oc. canadensis*, *Ps. howardii* and *Ur. lowi*.

As shown in Table 5, there were no significant differences in numbers caught in traps baited with R-octynol + CO_2 compared with the standard R : S octenol lure + CO_2. Both these baits yielded greater total mosquito collections and species collections, other than *Culex* spp. and *Cs. melanura*, than traps baited with S-octynol + CO_2 or CO_2 only. There were no significant differences between collections from traps baited with S-octynol + CO_2 and those baited with CO_2 only. Overall, this experiment suggests that R-octynol is as attractive as R : S racemic octenol, that S-octynol is not an attractant and that the most abundant *Culex* species are not attracted to these odours.

The sixth experiment, also with the Coleman traps, yielded a total of 12 031 ♀ and 15 ♂ mosquitoes, comprising 22 species over 16 nights (Table 6). Traps baited with R-octynol + CO_2 caught the greatest number (3418 ♀, 5 ♂), followed by R-octenol + CO_2 (3077 ♀, 4 ♂) and the standard R : S octenol lure + CO_2 (2918 ♀, 2 ♂). The R-octenol : R-octynol mix lure + CO_2 was least productive (2618 ♀, 4 ♂). In descending order of abundance, species were: 4120 ♀ *Oc. atlanticus*; 2389 ♀, 1 ♂ *An. crucians*; 2031 ♀ *Oc. infirmatus*; 1702 ♀ *Oc. canadensis*; 600 ♀ *Ps. ferox*; 571 ♀ *Cq. perturbans*; 283 ♀, 5 ♂ *Cx. salinarius*; 131 ♀ *Cx. erraticus*; 72 ♀, 4 ♂ *Cs. melanura*; 44 ♀ *Oc. triseriatus*; 25 ♀ *Oc. dupreii*; 19 ♀ *Cx. nigripalpus*; 15 ♀ *Ae. vexans*; 8 ♀ *An. quadrimaculatus*; 7 ♀ *Cx. quinquefasciatus*; 4 ♀ *Oc. mitchellae*; 2 ♀ *An. punctipennis*; 2 ♀ *Ae. albopictus*; 2 ♀, 5 ♂ *Cx. territans*; 2 ♀ *Or. signifera*; 1 ♀ *Ps. columbiae*, and 1 ♀ *Ma. titillans*. Species not collected with the standard R : S octenol lure were *Ae. albopictus* and *Ps. columbiae*; those not collected with R-octynol were *Cx. quinquefasciatus*, *Cx. territans*, *Oc. mitchellae* and *Ma. titillans*; those not collected with the R-octenol : R-octynol mix were *An. punctipennis*, *Ae. albopictus*, *Oc. mitchellae*, *Ma. titillans*, *Ps. columbiae* and *Or. signifera*; and those not collected with R-octenol + CO_2 were *An. punctipennis*, *Ma. titillans*, *Or. signifera* and *Cx. territans*. Traps baited with the R-octenol : R-octynol mix + CO_2 caught slightly fewer than those with the standard R : S octenol lure + CO_2, but there were no significant differences in catch between any of the treatments (Table 6), suggesting that there is little difference between the attractiveness of R-octenol and that of R-octynol.

Discussion

These data indicate that both R-octenol and R-octynol serve as attractants which enhance the activity of CO_2 for mosquitoes of several species, but not *Cs. melanura* or the most abundant *Culex* spp. Based on data obtained from preliminary experiments, we thought that octynol enantiomers might attract mosquito species that are attracted to octenol, while not repelling species such as *Cx. nigripalpus* that normally are not attracted to octenol (Kline, 1994). What we see from the data, however, is that the responses of mosquitoes to

Table 6. Mean catch (± standard errorSE) per trap-day* of most frequently caught species for different treatments of odour-baited Coleman MD-2500 propane-powered traps (traps produced ~ 500 mL/min CO_2, heat and water vapour), Gainesville, FL; four replicates of a 4 × 4 Latin square experiment; 28 April–18 May 2005.

	Attractant			
Species	R : S octenol	R-octynol	50 : 50 mix	R-octenol
Anopheles crucians	36.2 (7.16)	37.6 (8.61)	29.1 (4.23)	46.3 (7.74)
Culex salinarius	3.8 (0.84)	4.1 (1.15)	4.2 (1.08)	5.4 (1.45)
Culex erraticus	1.8 (0.44)	2.5 (0.74)	1.9 (0.46)	1.9 (0.43)
Culiseta melanura	1.6 (0.33)	1.4 (0.52)	1.0 (0.33)	1.1 (0.36)
Coquillettidia perturbans	7.5 (1.38)	8.4 (1.69)	9.1 (2.44)	10.5 (2.15)
Ochlerotatus atlanticus	65.0 (17.16)	68.8 (21.2)	56.9 (9.63)	66.6 (14.57)
Ochlerotatus canadensis	26.4 (5.88)	33.6 (10.52)	23.8 (4.21)	22.4 (3.91)
Ochlerotatus infirmatus	29.2 (9.77)	46.1 (27.98)	27.6 (7.83)	23.8 (5.52)
Psorophora ferox	9.5 (2.82)	8.1 (2.12)	8.1 (1.91)	11.3 (4.21)
Total	182.3 (40.53)	213.6 (70.39)	163.6 (21.52)	192.3 (32.77)

*$n = 16$ trap-days per treatment; means in the same row are not significantly different ($P > 0.05$); Tukey's means separation applied to log ($n + 1$) transformed data.

octenol and octynol seem to be similar. Equivalent enantiomers of both compounds seem to have similar patterns of mosquito attractancy: R-enantiomers appear to be better attractants than S-enantiomers, which are neutral or even repellent at the levels used in these experiments. Overall, these results support the findings of Kline (1994). The largest enhancement effect of the R-octenol and R-octynol enantiomers with CO_2 was with *An. crucians*. With the exception of Experiment 2, these enantiomers may have increased *An. crucians* collections four- to eight-fold compared with traps baited with only CO_2. Overall, R-octenol seemed to be the best treatment for most species, except in Experiment 6, where R-octynol was as effective as R-octenol. Generally, the results also confirm that the S-isomers of octenol and octynol are not attractive, and may even reduce the attractiveness of racemic mixtures.

These studies also support previous findings that octenol is not equally attractive to all species of mosquitoes. Specifically, octenol has not been demonstrated to be a strong attractant for *Stegomyia* mosquitoes. In our experiments, few *Ae. albopictus* were collected compared with other species. The majority of those caught were either in the CO_2-only traps or those baited with S-octynol + CO_2. In an Australian study, a comparison of octenol and CO_2 as attractants for *Ae. aegypti* (*Oc. aegypti*) with Fay-Prince traps in Queensland reported that octenol significantly decreased collections (Canyon & Hii, 1997). Similar results were reported recently with another *Stegomyia* mosquito, where significantly more *Ae. albopictus* were collected in Maryland with Fay-Prince traps baited with CO_2 or CO_2 + octenol, compared with unbaited or octenol-only baited traps, and octenol was found to be of no benefit (Shone *et al.*, 2003). The data reported by Russell (2004) on the relative catch sizes of *Aedes* (*Stegomyia*) species in the Pacific region with Center for Disease Control (CDC)-type miniature light traps or Encephalitis Virus Surveillance (EVS)-type traps baited with CO_2 and/or octenol indicate that CO_2 increases the catch in these traps, whereas octenol is not an attractant, either alone or in combination with CO_2.

No other studies have reported on the response of mosquitoes to the enantiomers of either octenol or octynol, and few studies with any other haematophagous insects have been conducted with these compounds. Hall *et al.* (1984) found that for tsetse species there were no differences in electroantennogram (EAG) activity, wind tunnel bioassay attractancy or attractiveness in the field between the two octenol enantiomers. Saini *et al.* (1989) also found that 1-octen-3-one and racemic octynol evoked EAG responses comparable with those evoked by racemic octenol for *G. morsitans morsitans*. A more recent report found that both commercially available octynol stereoisomers stimulated the antennae of *Glossina brevipalpis* (International Atomic Energy Agency, 2003).

Working with a non-haematophagous insect, Pierce *et al.* (1989) found that *O. mercator* grain beetles responded similarly to (R)-(−)-, (S)-(+)- and racemic 1-octen-3-ol when tested in a pitfall olfactometer, over an experimental stimulus range of 0.1 ng to 10 μg.

In contrast with these previous studies, we found a difference in the responses of mosquitoes to the R- and S-enantiomers of octenol and octynol; in both cases the R-enantiomer was attractive and the S-enantiomer did not increase CO_2-baited trap catches. Our findings are consistent with those for other semiochemicals that occur in isomeric forms; despite the similarity in structure, one enantiomer tends to be responsible for a particular effect, and the other is either neutral or involved with an entirely different biochemical reaction. However, we found that the closely related compounds, R-octenol and R-octynol, had a surprisingly similar effect on CO_2-baited trap catches, although of the two compounds, R-octenol generally had a greater effect. More research needs to be conducted with these enantiomers against a wider variety of mosquito species from different geographic regions and against other haematophagous insects to ascertain the differential efficacy of these odours. In particular, it would be interesting to explore the biochemistry of these chemicals in order to understand how they are detected by mosquito sensory systems and how their effect on the behaviour of mosquito species can vary to such a degree. Research on R-enantiomer lures alone and in combination with other kairomones would be especially interesting. Currently, synthetic octenol, which is a racemic mixture, is used in all commercially available lures. Although our data indicate that lures with the R-enantiomer alone may be more attractive, they would also be more expensive to produce. However, recent unpublished dose-response field studies conducted by Bedoukian Research Inc., indicate that far less (one-tenth as much) of the R-enantiomer of octenol needs to be incorporated in the lures to attain the same level of attractancy as the currently used commercial racemic octenol lures (R. H. Bedoukian, personal communication), although our findings did not show such a clear difference between R-octenol only and the racemic mixtures. This may be because all of our traps were also baited with CO_2.

Further studies should incorporate dose-response assays of various octenol and octynol enantiomer blends on their attractiveness to mosquitoes. Such efforts should enable the development of more effective generalized and specific lures for mosquitoes and other haematophagous insects.

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References

Atwood, D.W.R. & Meisch, M.V. (1993) Evaluation of 1-octen-3-ol and carbon dioxide as black fly (Diptera: Simuliidae) attractants in Arkansas. *Journal of the American Mosquito Control Association*, **9**, 143–146.

Becker, N., Zgonda, M., Petric, D. & Ludwig, M. (1995) Comparison of carbon dioxide, octenol and a host-odour as mosquito attractants in the Upper Rhine Valley, Germany. *Medical and Veterinary Entomology*, **9**, 377–380.

Buttery, G. & Kamm, J.A. (1980) Volatile components of alfalfa: possible insect host plant attractants. *Journal of Agriculture and Food Chemistry*, **28**, 978–981.

Canyon, D.V. & Hii, J.L.K. (1997) Efficacy of carbon dioxide, 1-octen-3-ol, and lactic acid in modified Fay-Prince traps as compared to man-landing catch of *Aedes aegypti*. *Journal of the American Mosquito Control Association*, **13**, 66–70.

Cheke, R.A. & Garms, R. (1987) Trials of attractants to enhance biconical trap catches of *Simulium yahense* and *Simulium sanctipauli s.l. Tropical Medical Parasitology*, **38**, 62–63.

Cork, A. (1994) Identification of electrophysiologically active compounds for New World screwworm, *Cochliomyia hominivorax*, in larval wound fluid. *Medical and Veterinary Entomology*, **8**, 151–159.

Cork, A. & Park, K.C. (1996) Identification of electrophysiologically active compounds for the malaria mosquito, *Anopheles gambiae*, in human sweat extracts. *Medical and Veterinary Entomology*, **10**, 269–276 (Erratum **11**, 104).

Darsie, R.F. & Ward, R.A. (2005) *Identification and Geographical Distribution of the Mosquitoes of North America, North of Mexico*, p. 384. University Press of Florida, Gainesville, FL.

Dijkstra, F.Y. & Wiken, T.O. (1976) Studies on mushroom flavours. I. Organoleptic significance of constituents of the cultivated mushroom, *Agaricus bisporous*. *Zeitschrift für Lebensmittel-Untersuchung und -Forschung A*, **160**, 255–262.

French, F.E. & Kline, D.L. (1989) 1-octen-3-ol, an effective attractant for Tabanidae (Diptera). *Journal of Medical Entomology*, **26**, 459–461.

Gibson, G. & Torr, S.J. (1999) Visual and olfactory responses of haematophagous diptera to host stimuli. *Medical and Veterinary Entomology*, **13**, 2–23.

Hall, D.R., Beevor, P.S., Cork, A., Nesbitt, B.F. & Vale, G.A. (1984) 1-Octen-3-ol. A potent olfactory stimulant and attractant for tsetse isolated from cattle odours. *Insect Science and Applicata*, **5**, 335–339.

Hayes, R.O., Doane, O.W., Sakolsky, G. & Benick, S. (1993) Evolution of attractants in traps for greenhead fly (Diptera: Tabanidae) collections on Cape Cod, Massachusetts salt marsh. *Journal of the American Mosquito Control Association*, **9**, 436–440.

Honkanen, E. & Moisio, T. (1963) On the occurrence of oct-1-en-3-ol in clover plants. *Acta Chemica Scandinavica*, **17**, 858.

International Atomic Energy Agency (2003) *Improved Attractants for Enhancing Tsetse Fly Suppression*, p. 121. IAEA-TECDOC-1373. IAEA, Vienna, Austria.

Kaminski, E., Stawicki, S., Wasowicz, E. & Przylski, R. (1973) Detection of deterioration of grain by gas chromatography. *Annales de Technologie Agricole*, **22**, 401–407.

Kaminski, E., Stawicki, S. & Wasowicz, E. (1974) Volatile flavour compounds produced by moulds of *Aspergillus*, *Penicillium*, and *Fungi imperfecti*. *Applied Microbiology*, **27**, 1001–1004.

Kemme, J.A., Van Essen, P.H.A., Ritchie, S.A. & Kay, B.H. (1993) The response of mosquitoes to CO_2 and 1-octen-3-ol in South East Queensland, Australia. *Journal of the American Mosquito Control Association*, **9**, 431–435.

Kline, D.L. (1994) Olfactory attractants for mosquito surveillance and control: 1-octen-3-ol. *Journal of the American Mosquito Control Association*, **10**, 280–287.

Kline, D.L. (1999) Comparison of two American Biophysics mosquito traps: the Professional and a new counterflow geometry trap. *Journal of the American Mosquito Control Association*, **15**, 276–282.

Kline, D.L., Dame, D.A. & Meisch, M.V. (1991a) Evaluation of 1-octen-3-ol and carbon dioxide as attractants for mosquitoes associated with irrigated rice fields in Arkansas. *Journal of the American Mosquito Control Association*, **7**, 165–169.

Kline, D.L., Hagan, D.V. & Wood, J.R. (1994) *Culicoides* responses to 1-octen-3-ol and carbon dioxide in salt marshes near Sea Island, Georgia, U.S.A. *Medical and Veterinary Entomology*, **8**, 25–30.

Kline, D.L., Takken, W., Wood, J.R. & Carlson, D.A. (1990a) Field studies on the potential of butanone, carbon dioxide, honey extract, 1-octen-3-ol, l-lactic acid and phenols as attractants for mosquitoes. *Medical and Veterinary Entomology*, **4**, 383–391.

Kline, D.L., Wood, J.R. & Cornell, J.A. (1991b) Interactive effects of 1-octen-3-ol and carbon dioxide on mosquito (Diptera: Culicidae) surveillance and control. *Journal of Medical Entomology*, **28**, 254–258.

Kline, D.L., Wood, J.R. & Morris, C.D. (1990b) Evaluation of 1-octen-3-ol as an attractant for *Coquillettidia perturbans*, *Mansonia* spp. and *Culex* spp. associated with phosphate mining operations. *Journal of the American Mosquito Control Association*, **6**, 605–611.

Pierce, A.M., Pierce, H.D., Borden, J.H. & Oehlschlager, A.C. (1989) Production dynamics of cucujolide pheromones and identification of

1-octen-3-ol as a new aggregation pheromone for *Oryzaephilus surinamensis* and *O. mercator*. *Environmental Entomology*, **18**, 747–755.

Raymer, J., Wiesler, D., Novotny, M., Asa, C., Seal, U.S. & Mech, L.D. (1985) Chemical investigations of wolf (*Canis lupus*) anal-sac secretion in relation to breeding season. *Journal of Chemical Ecology*, **11**, 593–608.

Russell, R.C. (2004) The relative attractiveness of carbon dioxide and octenol in CDC- and EVS-type light traps for sampling the mosquitoes *Aedes aegypti* (L.), *Aedes polynesiensis* Marks, and *Culex quinquefasciatus* Say in Moorea, French Polynesia. *Journal of Vector Ecology*, **29**, 309–314.

Saini, R.K., Hassanali, A. & Dransfield, R.D. (1989) Antennal responses of tsetse to analogues of the attractant 1-octen-3-ol. *Physiological Entomology*, **14**, 85–90.

Sant'Ana, A.L., Eiras, A.E. & Cavalcante, R.R. (2002) Electroantennographic responses of the *Lutzomyia (Lutzomyia) longipalpis* (Lutz & Neiva) (Diptera: Psychodidae) to 1-octen-3-ol. *Neotropical Entomology*, **31**, 13–17.

SAS Institute (2001) *SAS/STAT User's Manual, Version 8.2*. SAS Institute, Cary, NC.

Shone, S.M., Ferrano, P.N., Lesser, C.R., Glass, G.E. & Norris, D.E. (2003) Evaluation of carbon dioxide- and 1-octen-3-ol-baited Centers for Disease Control Fay-Prince traps to collect *Aedes albopictus*. *Journal of the American Mosquito Control Association*, **19**, 445–447.

Takken, W. & Kline, D.L. (1989) Carbon dioxide and 1-octen-3-ol as mosquito attractants. *Journal of the American Mosquito Control Association*, **5**, 311–316.

Van Essen, P.A., Kemme, J.A., Ritchie, S.A. & Kay, B.H. (1994) Differential responses of *Aedes* and *Culex* mosquitoes to octenol or light in combination with carbon dioxide in Queensland, Australia. *Medical and Veterinary Entomology*, **8**, 63–67.

Wurzenberger, M. & Grosch, W. (1982) The enzymatic oxidative breakdown of linoleic acid in mushrooms (*Psalliota bispora*). *Zeitschrift fur Lebensmittel-Untersuchung und-Forschung*, **175**, 186–190.

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